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Clearance and Metabolism of Starch Foods in the Oral Cavity

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Key Words
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Abstract

The presence of carbohydrates and organic acids was monitored in the oral cavity over a 3-hour period following the ingestion of six foods containing cooked starch (popcorn, potato chips, corn flakes, bread stick, hard pretzel and wheat cracker) and compared to a food containing sugar (chocolate-covered candy bar). Oral fluid samples were collected at 30-min intervals from five different tooth sites from 7 volunteers using absorbent paper points. Samples were analyzed for carbohydrates and organic acids using high-performance liquid chromatography. Analytical data for each food were pooled and compared to the results of the sugar food. The amount of lactic acid produced 30 min after ingestion was highest with the potato chips and lowest with the corn flakes. Potato starch contributed more readily to oral lactic acid production than wheat or corn starch. A direct linear relationship existed between lactic acid production and the presence of oral glucose produced from starch, which occurred via the metabolites maltotriose and maltose. Oral clearance of foods containing cooked starch proceeded significantly slower than that of the sugar food, thus contributing to a prolonged period of lactic acid production.

Introduction

Testing the potential cariogenicity of foods often relies on plaque pH measurement [1, 2]. Inherent properties of the foods, such as fermentable carbohydrates, buffering capacity, solubility and induction of salivary stimulation, could interfere with such an evaluation [3]. Most studies on plaque acid production following ingestion of certain foods by human subjects were short-term studies examining only the first 30 min after ingestion [4, 5]. Recently, a high-performance liquid chroma-

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tography (HPLC) method has been utilized [6] to follow the oral food clearance and intra-oral acid production of various foods qualitatively and quantitatively for up to 3 h in situ [7]. This HPLC technique proved to be useful in the evaluation of oral clearance of different foods containing various carbohydrates such as sugars and/or cooked starch. While the cariogenic potential of sugars is generally accepted and adequately studied, several authors have lately recognized that cooked starch has a significant cariogenic potential as well [2, 8–12]. Some of these studies were limited with regard to in vitro measurement of acid formation or measurement of pH changes or use of animal models. This clinical oral clearance study intended to explore the chronology of the oral catabolism of cooked starch to fermentable glucose and its subsequent conversion into lactic acid by plaque bacteria in situ.

Materials and Methods

Test Foods

Eight different foods were tested in 7 volunteers: a chocolate-covered candy bar (Milky Way; Mars, Hackettstown, N.J., USA), popcorn (Wise Foods, Borden, Columbus, Ohio, USA), a whole wheat cracker (Nabisco Brands, East Hanover, N.J., USA), a bread stick (Stella d'Oro Biscuit, Bronx, N.Y., USA), corn flakes (Kellog USA, Battle Creek, Mich., USA), a hard pretzel (The Bachman Company, Reading, Pa., USA) and potato chips (Wise Foods, Borden).

Collecting of Clinical Specimens

Seven volunteer healthy dental students were advised to refrain from food intake for 2 h before the beginning of the initial test. These volunteers were not preconditioned, i.e., they received no instructions on oral hygiene and eating habits before the experiment. The presence of a small amount of dental plaque was desirable for subsequent optimum starch food conversion. They were offered the test foods in single portions of 25 g. The volunteers had a normal salivary flow; subjects with strongly decreased salivary flow were excluded. However, their eating time varied (swallowing bolus); this phenomenon was discussed earlier [6]. Besides a few amalgam fillings, the volunteers had no excessive restorations, such as crowns, partial and full dentures, which could have affected and falsified oral fluid collection. The research protocol using human subjects was reviewed and approved by the appropriate institutional committee. The subjects were not allowed to take in fluids during the experiment.

Following ingestion, oral fluid samples were taken from five tooth sites, using one sterile paper point (No. 2717; Johnson and Johnson, New Brunswick, N.J., USA) for each site, i.e. the interproximal surface of the last two teeth in the lower right quadrant, the interproximal surface of the last two teeth of the upper left quadrant, the occlusal surface of the last tooth in the lower left quadrant, the occlusal surface of the last tooth in the upper right quadrant, and the lingual surface of the last tooth in the lower right quadrant. Sampling was conducted at time 0, immediately before eating the test food (baseline) and at 30, 60, 90, 120, 150 and 180 min after ingestion of food. Vials with the five pooled paper points from each individual for each sampling time were kept individually frozen at –80 °C until extraction.

Extraction of Carbohydrates and Organic Acids

1.0 ml of distilled water was added to five pooled paper points (containing on average 60 mg of combined oral fluid); the vial was shaken for 10 s, and again for 10 s after a 20-min time period. The supernatant was filtered through a 0.2-µm membrane filter (Acro LC13; Gelman Sciences, Ann Arbor, Mich., USA), and then 80-µl portions were analyzed using HPLC. The composition and chemical stability of these samples were discussed earlier [6].

HPLC Analysis

A liquid chromatographic system composed of an ion exclusion column (Aminex HPX-87H; Bio-Rad Laboratories, Richmond, Calif., USA) and a refractive index detector (Shodex RI SE61; Showa Denko America, New York, N.Y., USA) was used as described before [7].

Statistics

Data were analyzed using the statistical DOS package SPSS/PC+, Version 4.0 (SPSS, Chicago, Ill., USA). Subjective ratings were compared by analysis of variance. Lactic acid data of the different foods were analyzed by one-way analysis (ranges for the 0.05 level) using the Tukey-HSD procedure (statistical results see Discussion).

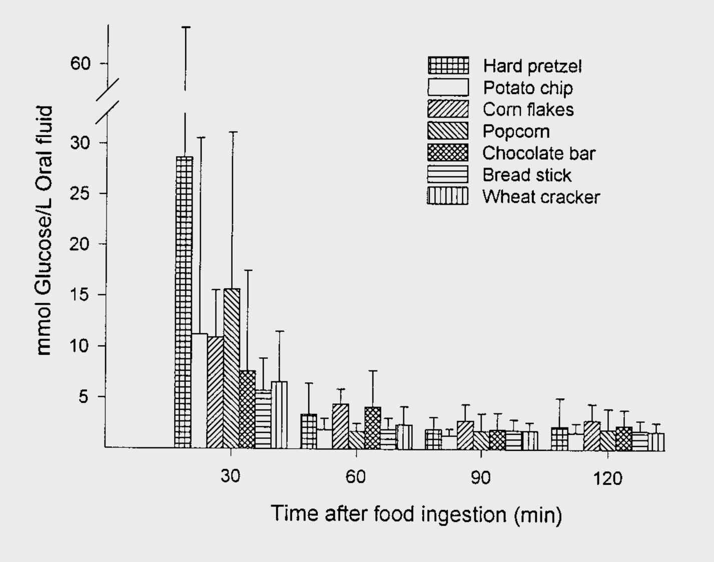

Fig. 1. Average concentrations of glucose in oral fluid after eating various foods measured at 30-min intervals for a time period of 120 min (averaged from all volunteers).

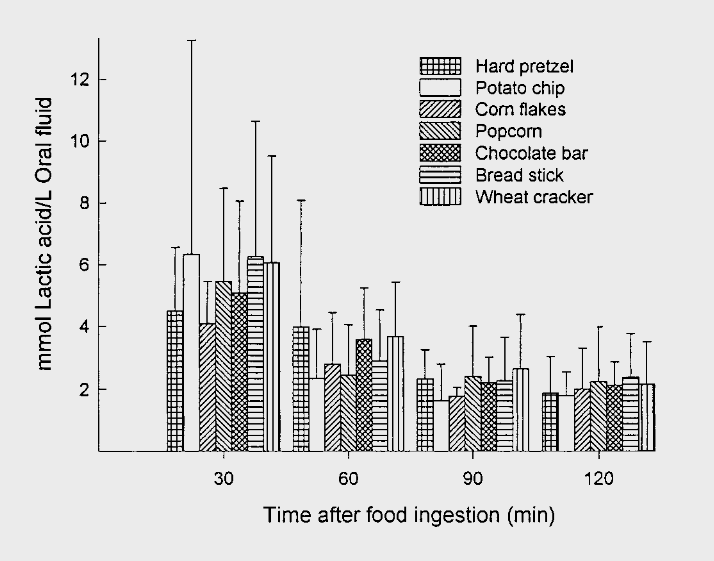

Fig. 2. Average lactic acid concentrations in oral fluid after eating various foods measured at 30-min intervals for a time period of 120 min (averaged from all volunteers).

Results

Chocolate-Covered Candy Bar

Glucose clearance proceeded in a normal and predictable pattern (fig. 1). The highest carbohydrate concentration – as expected – was found immediately after ingestion (30 min). Carbohydrates had almost completely cleared 90 min after ingestion. Lactic acid concentrations were highest between 30 and 60 min, and decreased subsequently to relative baseline within 2 h (fig. 2).

Table 1. Total concentrations of maltotriose and maltose derived from various starch foods during 2 h of oral clearance[1]

Food		Clearance time, min	Maltotriose, mmol/l oral fluid	Maltose, mmol/l oral fluid
Hard pretzel	during	180	0.486 ± 0.849	2.079 ± 4.164
	after	30	1.822 ± 1.245	7.962 ± 9.586
		60	0.755 ± 0.717	1.226 ± 0.634
		90	0.392 ± 0.857	1.016 ± 0.423
		120	0.039 ± 0.056	1.136 ± 0.451
Potato chip[2]	during	180	0.051 ± 0.093	1.346 ± 0.945
	after	30	0.149 ± 0.147	1.879 ± 1.795
		60	0.029 ± 0.049	1.207 ± 0.618
		90	0.036 ± 0.073	1.145 ± 0.584
		120	0.027 ± 0.054	1.593 ± 1.089
Corn flakes	during	180	0.543 ± 0.492	1.958 ± 2.179
	after	30	0.700 ± 0.466	3.785 ± 5.227
		60	0.435 ± 0.254	1.304 ± 0.266
		90	0.565 ± 0.493	1.672 ± 1.023
		120	0.520 ± 0.482	2.161 ± 1.522
Popcorn[2]	during	180	0.837 ± 0.746	1.717 ± 3.917
	after	30	1.874 ± 1.239	6.969 ± 9.098
		60	0.649 ± 0.263	0.726 ± 0.424
		90	0.821 ± 0.700	0.974 ± 0.754
		120	0.769 ± 0.425	0.914 ± 0.753
Bread stick[2]	during	180	1.539 ± 1.539	1.905 ± 2.732
	after	30	3.118 ± 3.452	6.006 ± 5.473
		60	1.293 ± 0.747	1.122 ± 0.706
		90	1.344 ± 0.752	1.192 ± 0.695
		120	1.356 ± 0.792	1.201 ± 0.736
Wheat cracker[2]	during	180	1.372 ± 0.942	1.547 ± 1.922
	after	30	2.346 ± 1.482	4.202 ± 4.141
		60	1.388 ± 0.849	1.275 ± 0.704
		90	1.381 ± 0.824	1.126 ± 0.621
		120	1.244 ± 0.762	1.125 ± 0.728

[1] Individual data at 150 and 180 min not shown.
[2] Tested on 7 volunteers; all other foods, on 6 volunteers.

Potato Chip

30 min after ingestion, high concentrations of maltose and maltotriose (di- and trisaccharides, respectively) were observed (table 1). These polysaccharides were rapidly hydrolyzed into glucose. Maltotriose was completely degraded within 60 min. The glucose concentration was relatively high after 120 min (fig. 1). This observation was confirmed by a steady lactic acid level in all samples af-

ter ingestion. The lactic acid concentration was somewhat higher at 120 than at 90 min (fig. 2).

Wheat Cracker

Mono-, di- and trisaccharides were detected 30 min after ingestion. Di- and trisaccharide concentrations were highest 30 min after ingestion. After 60 min, maltotriose, maltose and glucose concentrations remained constant but were significantly elevated for up to 2 h (table 1; fig. 1). Lactic acid concentrations reached an optimum after 30 min and slowly decreased toward the 2-hour mark. At 120 min, the observed lactic acid concentration was slightly higher than that detected with the chocolate-covered candy bar (fig. 2). The wheat cracker was the starch-containing food that produced the highest quantity of acid (table 2).

Popcorn

Highest concentrations of maltotriose, maltose and glucose were found at 30 min (table 1; fig. 1). After initial clearance, the glucose concentration increased slightly up to 120 min (fig. 1). The lactic acid concentration decreased slowly throughout the experiment. At 120 min, the starch foods popcorn and bread stick generated the highest acid concentrations (fig. 2).

Corn Flakes

Carbohydrate clearance progressed consistently. The concentration of maltotriose, maltose and glucose peaked at 30 min (table 1; fig. 1). The carbohydrate level at 120 min was approximately 25% of that at 30 min (fig. 1). The lactic acid concentration steadily declined over 90 min. At 120 min, the lactic acid concentration was slightly lower than with the sugar food (fig. 2).

Table 2. Average concentrations of lactic acid derived from various foods during 3 h of oral clearance[1]

Food	Clearance time, min	Lactic acid mmol/l oral fluid
Hard pretzel	30–180	2.613 ± 2.292
Potato chip[2]	30–180	2.430 ± 3.190
Corn flakes	30–180	2.142 ± 1.421
Popcorn[2]	30–180	2.596 ± 2.083
Chocolate bar	30–180	2.816 ± 1.751
Bread stick[2]	30–180	2.933 ± 2.456
Wheat cracker[2]	30–180	3.064 ± 2.399

[1] Averaged from individual data at sampling points of 30, 60, 90, 120, 150 and 180 min.

[2] Tested on 7 volunteers; all other foods, on 6 volunteers.

Bread Stick

Carbohydrate clearance progressed slowly. At 30 min, an unusually high concentration of maltose was observed (table 1). The lactic acid concentration dropped steadily over 120 min and remained at 25% of that of the 30-min level. That level was significantly higher than with the sugar product (fig. 2).

Hard Pretzel

Carbohydrate clearance (table 1; fig. 1) and acid production (fig. 2) followed a pattern similar to that of the bread stick. At 30 min, extremely high concentrations of maltotriose, maltose and glucose were observed. This starch food produced the highest amount of glucose for the duration of the experiment (table 3). At 60 and 90 min, lactic acid concentrations were found to be somewhat higher than with the chocolate-covered candy bar (fig. 2).

The baseline data obtained and averaged from 46 volunteers are summarized in table 4. The concentration of glucose was found to be 1.897 mmol/l oral fluid. There was always a

Table 3. Average concentrations of glucose derived from various foods during 3 h of oral clearance[1]

Food	Clearance time, min	Glucose mmol/l oral fluid
Hard pretzel	30–180	6.209 ± 15.014
Potato chip[2]	30–180	2.959 ± 7.686
Corn flakes	30–180	4.002 ± 3.725
Popcorn[2]	30–180	3.755 ± 7.484
Chocolate bar	30–180	3.919 ± 5.058
Bread stick[2]	30–180	2.390 ± 2.024
Wheat cracker[2]	30–180	2.443 ± 2.598

[1] Averaged from individual data at sampling points of 30, 60, 90, 120, 150 and 180 min.
[2] Tested on 7 volunteers; all other foods, on 6 volunteers.

Table 4. Baseline data of carbohydrates and acids in oral fluid before food ingestion (mean from 46 volunteers)

Compound	Carbohydrates/acids mmol/l oral fluid
Maltotriose	0.497 ± 0.598
Maltose	1.070 ± 0.888
Sucrose	–[1]
Lactose	–[1]
Glucose	1.897 ± 1.364
Galactose	–[1]
Fructose	0.466 ± 0.300
Lactic acid	2.046 ± 1.541
Formic acid	5.420 ± 5.851
Acetic acid	9.102 ± 9.270
Propionic acid	0.649 ± 0.539
Ethanol	–[1]
n-Butyric acid	–[1]

All baseline data from this study averaged (from 6 to 7 volunteers and the 7 food groups tested).
[1] Compound not analyzed in this series.

small amount of lactic acid (2.046 mmol/l oral fluid). Formic and acetic acid were consistently found in these samples (on average 5.420 and 9.102 mmol/l oral fluid, respectively). Baseline concentrations of the latter two acids were found to be surprisingly high, but external contamination of the samples could be excluded. However, after food intake the concentrations of formic and acetic acid dropped to an insignificant level below 5% of total acid production (data not shown).

Discussion

In order to obtain a representative oral fluid specimen for this study, samples from five different oral sites were pooled. Some sugars stimulate salivary flow at a different rate, which may affect the rate of food clearance in the oral cavity. Unstimulated saliva does not mix well in the mouth, and the buffering capacity of saliva from the different glands varies significantly [13]. Other investigators have reported that glucose clearance varies at the different sites in the oral cavity [14]. No foodstuff can be considered a 'pure' food. Most sweet foods are composed of a mixture of sugars which are very often supplemented with starches and other compounds. We observed a significant difference in salivary flow rate among the volunteers (data not shown). It was noticed that ingestion time (swallowing bolus) of the same test food varied among the individual volunteers from 4 to 16 min (data not shown). A lower salivary flow, bordering on the condition in a 'dry mouth' patient, resulted in a delay in peak lactic acid production. This is also reflected in the higher standard deviations of the obtained lactic acid data (fig. 2).

Small amounts of glucose were always found in oral fluid at baseline before ingestion of the test foods (table 4). Other investigators found glucose (2.22 mmol/l) in whole saliva

[15] and detected glucose (5–25 μmol/l) in parotid saliva before carbohydrate intake [16]. The observed concentrations of formic and acetic acid were unexpectedly high at baseline levels (table 4). Other investigators found similar concentrations of organic acids in starved plaque fluid [17, 18]. These acids could have originated either from the metabolism of carbohydrates in food remnants or from soluble polysaccharides stored or retained in plaque, such as levan and starch. After ingestion of the test foods, formic and acetic acid concentrations dropped (data not shown), whereas lactic acid levels increased significantly (fig. 2). Two hours after food ingestion, lactic acid levels approached baseline levels again (fig. 2); for this reason, clearance data beyond 120 min (sampling points at 150 and 180 min) are not reported and will not be discussed here. Active carious dentin contains up to 88.2% of lactate. In arrested lesions or carious dentin situated beneath restorations, acetate and propionate appear dominant [19]. However, the concentrations of formic, acetic and propionic acid are not described here since they contributed only ≤ 5% to the total amount of acids produced during long-term clearance. Similar findings have also been reported elsewhere [20–24].

Significant differences in lactic acid production were observed among the various foods tested. The predominantly starch-containing foods (e.g., wheat cracker) generated significantly more lactic acid during 120 min than the food containing carbohydrate sweeteners only (chocolate-covered candy bar). Data of total lactic acid production for the 180-min duration of the experiment indicated that the wheat cracker produced the highest concentration of lactic acid, whereas the corn flakes produced the lowest concentration (table 2).

Retention times of different foods profoundly affect bacterial acid production [25]. Foods containing starch are not readily soluble in saliva; they take longer to clear than the foods containing sugars only. Foods containing mono- and disaccharides but no starch clear the oral cavity more rapidly; maximum acid production occurs within 30 min after ingestion [7]. After 30 min, the concentration of acids diminishes rapidly, following the classical Stephan curve [26]. One-way analysis using the multiple-range test (Tukey-HSD procedure) on the variables ‘lactic acid’ by ‘time’ produced significant differences (ranges for the 0.05 level) for all tested foods at clearance times of 30 vs. 60 min, 30 vs. 90 min and 30 vs. 120 min. All of these discussed factors must be considered for a meaningful clinical interpretation of the obtained results which clearly indicate that starch-containing foods gave rise to significant amounts of glucose 30 min after ingestion (fig. 1). Apparently, the production of glucose occurs by the action of α-amylase on cooked starch via the intermediate degradation products maltotriose and maltose [27–30], which was also observed for the first time in this in situ study (table 1). It has been suggested that gelatinized starch granules, generally obtained by cooking or heat-processing of starches, are available for degradation by salivary α-amylase [31, 32]. Depending on the degree of food processing, gelatinized starches may influence their dental properties [11]. While some investigators believe that starch is broken down in the oral cavity by the action of salivary α-amylase [28], others also emphasize the importance of bacterial α-amylase in this process [27].

Analysis of the starch food clearance data yielded the following pattern for intra-oral starch breakdown: starch → maltotriose → maltose → glucose (table 1; fig. 1). Moreover, it was found that different types of starch (corn, potato or wheat starch) are metabolized at different rates in the oral cavity. This can be seen from the different concentrations of maltotriose vs. maltose obtained from the three types of starch tested during oral clearance (table 1). Potato and wheat starch gave rise to higher amounts of lactic acid than corn starch which is metabolized at a much slower rate (table 2; fig. 2). During testing of the demineralization ability of several starch foods [33], it was found that wholemeal bread and bran flakes were associated with much less demineralization than cooked rice and white bread. It appears that the carbohydrates of the more processed foods are readily available to the acidogenic plaque bacteria. Refined white flour (wheat germ and bran removed) was found to be more acidogenic than unrefined graham flour (containing both wheat germ and bran) in a decalcification study using bovine enamel slabs [34]. In a study using albino rats [35], a wheat starch diet produced a higher caries score (15.5) than a maize starch diet (12.75). Other investigators observed that fermentable maltose, produced from cooked wheat starch, contributed – via bacterial acid production – to significant intra-oral demineralization, whereas under the same conditions raw starch normally resulted in minor demineralization [9].

The most important finding of both this study and a previous investigation [7] is the demonstration of a marked difference between the oral clearance of foods containing primarily carbohydrate sweeteners and those containing primarily cooked starch. While acid production from sugar-containing foods rapidly diminishes over a 1-hour time period, acid production from the primarily starch-containing foods continues steadily beyond 2 h. A similar finding has been reported by other investigators [25], indicating that starch-containing foods appear to be more retentive than foods primarily containing sugars. The observed clearance data support the contention that foods containing cooked starch (slow clearance) can be labeled as 'sticky', whereas foods primarily containing sugars (fast clearance) can be labeled as 'non-sticky'.

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